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ABSTRACT

This study investigates the effect of a three-month physical fitness program on serum-free cholesterol concentration in four age and fitness adult groups. Twenty-four men were divided into the following groups: (a) young, low-fit; (b) old, low-fit; (c) young, high-fit; and (d) old, high-fit. All subjects had normal resting glucose and triglyceride levels and participated in a physical fitness program consisting of calisthenics, jogging, and recreational activities. Subjects were tested on a treadmill at the beginning and at the end of the three-month program, and venous blood samples were drawn at four stages of metabolic stress: rest, submaximal, maximal, and recovery. The analysis of variance technique was used to determine significant differences between pre- and post-tests, fitness levels, age groups, different metabolic stress levels, and the significant interactions of various factors. Data suggest the following conclusions: (a) there is no significant change in serum-free cholesterol level due to a three-month physical fitness program; (b) the high-fit group had a significantly lower free cholesterol level than the low-fit group did; (c) there was no significant difference between fitness groups for the percent-free cholesterol; (d) there were no significant differences between age groups for the free cholesterol or percent-free cholesterol; and (e) acute physical exercise increases the absolute and percent-free cholesterol from the resting state through the maximum exercise state. (PD)

THE EFFECT OF A THREE-MONTH PHYSICAL FITNESS PROGRAM ON SERUM FREE CHOLESTEROL*

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INTRODUCTION

Arteriosclerosis is the major cause of death in the United States (21). The association of hyperlipidemia with the arteriosclerotic process is well documented in a wide range of experimental animals (3) and in humans (4). Hyperlipidemia is a risk factor in arteriosclerosis which is susceptible to alteration. Data from large experiments like the Framingham Study (10) reveal that individuals in the upper 25% in terms of serum cholesterol concentration have a much higher risk of myocardial infarction than do individuals in the lower 25%. The risk of myocardial infarction increased in proportion to the serum cholesterol concentration from the lowest to highest values.

Man has a high concentration of total cholesterol compared to other species (5). Approximately 80-90% of the total cholesterol of most tissue consists of free cholesterol. The serum is an exception with only 20 to 33% in the form of free cholesterol (8). The remainder is esterfied to fatty acids.

In man, the lecithin-cholesterol acyltransferose (LCAT) enzyme catalyzes the transfer of fatty acids from the B-position of lecithin to free cholesterol in the plasma (16,18,19). Since human plasma does not contain enzymes that hydrolyze cholesterol esters, the esterification of free cholesterol represents the activity of LCAT. The breakdown of cholesterol esters probably occurs mainly in the liver (16).

It has been hypothesized that LCAT may be involved in preventing atherosclerosis since free cholesterol is more atherogenic than cholesterol ester (12). Soloff et al (22) indicated that men with myocardial infarction had a lower efficiency of cholesterol esterification per minute compared to healthy individuals.

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This study was an attempt to observe the effect of an exercise program on the serum free cholesterol level in four age and fitness groups. Since free cholesterol is more atherogenic than cholesterol ester, it is hypothesized that any treatment that would lower the serum free cholesterol would reduce the susceptibility to atherosclerosis.

Purpose of the Study: To determine the effect of a three-month physical fitness program on serum free cholesterol concentration in four age and fitness adult groups.

Procedures: Twenty-four men were divided into four equal groups (n=6) using the test criterion of Ismail et al (9), namely (I) a young, low-fit group; (II) an old low-fit group; (III) a young high-fit group, and (IV) an old high-fit group. The subjects were selected from 100 men who participated in the physical fitness program at Purdue University. The physical fitness program consisted of calisthenics, jogging and recreational activities such as basketball, volleyball, squash, and handball. In addition, all subjects had normal resting glucose and triglyceride levels.

The subjects were tested on a treadmill at the beginning and at the end of the three-month physical fitness program. Venous blood samples were drawn at four stages of metabolic stress: rest, submaximal, maximal and recovery. On the post test, two blood samples were drawn during maximal exercise, with the first blood sample taken at the same stress level that corresponded to the pre test and the second blood sample was obtained to reflect the new maximal level.

The biochemical variables were measured by colorimetric methods. The total lipid was measured according to Zollner and Kirsch (24). The triglyceride level was measured according to Oxford Laboratories Procedure (20). The glucose level was measured according to a Harleco Procedure (6). The total cholesterol and free cholesterol concentrations were measured by the Hycel Method (8).

The analysis of variance technique was used to determine if any significant differences between pre and post tests, fitness levels, age groups, different metabolic stress levels, and the significant interactions of the various factors. The analysis of variance design was a factorial with subjects being nested within groups.

RESULTS

Total Lipid and Total Cholesterol

The means and standard deviations for the variables: age, weight, physical fitness score, resting serum glucose and resting serum triglyceride, for the four groups are presented in Table 1. These variables were used to select the four groups. Regardless of physical fitness, the mean age of the old group was 52.9 years and the mean age of the young group was 36.7 years. Regardless of age, the mean weight of the low-fit groups at the pre test was 176.6 pounds and their mean physical fitness score was 296.3 while the high-fit group had mean values of 171.3 pounds and 404.9 respectively. There was a slight and insignificant decrease in weight from pre to post test and a significant increase in physical fitness score in all groups. Clinically all subjects were within the normal range of 70 to 120 mg% (6) for resting serum glucose level. The four group means were similar. There was a significant difference in resting serum triglyceride level between the high-fit and low-fit groups. The mean serum triglyceride levels were within the normal range of 30 to 200 mg% (20) from the clinical point of view.

Table 2 presents the means and standard deviations of the serum total lipid and the serum total cholesterol levels for the total group on the pre and post tests at different metabolic stress levels namely, rest, submaximal, maximal I, maximal II, and recovery. A pattern is shown for both the total lipid and cholesterol levels which starts with a resting mean value which increases during submaximal exercise, followed by a further increase during maximal exercise, and decreases during the 15 minute recovery period.

The total group on the pre test started with a total lipid level at rest of 586.1 mg%, which increased to 605.5 mg% during the submaximal exercise, then increased further to 781.8 mg% during maximal exercise, and decreased to 672.3 mg% during the recovery period. On the post test, the total lipid level was 601.3 mg% at rest, 624.8 mg% during submaximal exercise, 784.6 mg% at Max I, 833.5 mg% at Max II, and decreased to 720.5 mg% during the recovery period. These results are presented graphically in Figure 1. The four sub-groups had similar patterns as the total group. (See Figure 3 and Table 6).

The total group on the pre test started with cholesterol level of 236.8 mg% at rest, increased to 246.3 mg% during the submaximal exercise, then increased further to 271.2 mg% during the maximal exercise, and decreased to 239.5 mg% during the recovery period. On the post test, the total cholesterol level was 232.0 mg% at rest, 250.1 mg% during submaximal exercise, 273.5 mg% at Max I,

276.3 mg% at Max II, and decreased to 245.3 mg% during the recovery period. The four sub-groups had similar patterns as the total group. (See Figure 3 and Table 6).

The ANOVA results in Table 3 showed the differences between groups, tests, metabolic states and their interactions. The results indicated fitness groups, levels of stress and fitness by stress interaction were significant relative to total lipid. As to total cholesterol it was found that stress level, fitness by stress interaction, and time by stress interaction were significant.

Free Cholesterol

The means and standard deviations of the free cholesterol level and the percent free cholesterol for the total group on the pre and post tests are presented in Table 4. Again, a pattern is shown for both free cholesterol and the percent free cholesterol which starts with a resting mean value which increases during submaximal exercise, followed by a further increase during maximal exercise, and decreases during the recovery period. The percent free cholesterol values are given in parenthesis. They were obtained by:

$$\frac{\text{Free Cholesterol X 100}}{\text{Total Cholesterol}} = \% \text{ Free Cholesterol}$$

For the total group on the pre test, the free cholesterol level increased from 56.6 mg% (24.0%) at rest, to 60.9 mg% (24.8%) during submaximal exercise, to 68.6 mg% (25.4%) during maximal exercise, and decreased to 59.9 mg% (24.9%) during the recovery period. On the post test, the free cholesterol level for the total group increased from 53.9 mg% (23.1%) at rest, to 60.5 mg% (24.1%) during submaximal exercise, to 67.4 mg% (24.6%) during Max I and 67.3 mg% (24.4%) during Max II, with a decrease to 58.7 mg% (23.9%) during recovery period. The results for the total group are presented graphically in Figure 2. The four sub-groups had similar patterns as the total group (See Figure 4 and Table 7).

The ANOVA results in Table 5 showed the differences between groups, tests, metabolic states, and their interactions. The results indicated that fitness groups and levels of stress were significant relative to free cholesterol. The high-fit group had a significantly lower free cholesterol level than the low-fit group. However, when the percent free cholesterol level is observed, there was no significant difference between fitness groups. There was no significant difference

between age groups nor between the pre and post tests for either the free cholesterol level or the percent free cholesterol level.

There are highly significant differences among metabolic stress levels for both the free cholesterol and percent free cholesterol. The effect of the different metabolic states for the four groups on the pre and post tests can be seen in Figure 2. Again, the pattern is the same in regards to stress levels.

DISCUSSION

The major purpose of this study was to examine the effects of a three-month exercise program on the serum free cholesterol level. The findings indicated that there were no significant change in serum cholesterol level between the pre and post tests. Other studies (7,11) have found no significant reduction in total cholesterol due to exercise while the subjects were on a normal diet. Two studies (16,23) have noted that it was necessary to decrease the body weight to lower the serum cholesterol level. In this study, there was no significant loss in body weight. Chaikoff et al (2) have shown that the side chain of labeled cholesterol is oxidized to CO_2 during exercise, however the labeled carbon on the ring cholesterol is not oxidized. Furtnermore, the effects of the program on different levels of fitness and age groups on serum free cholesterol were examined. The results indicated that the high-fit group had a significantly lower serum free cholesterol level than did the low-fit group. This could be related to the percent body fat as the high-fit group were leaner than the low-fit group. Montoye et al (14) found a significant positive correlation between total cholesterol and body fatness (sum of four skinfolds) in 4,000 subjects aged 10 to 64 years. When the percent free cholesterol was examined, no significant differences occured. Our results showed no significant differences between young and old exercise groups for either free cholesterol level or percent free cholesterol level. Other results (14) have indicated that there is an increase in the proportion of free cholesterol with age.

The effect of the acute exercise was to increase both the absolute free cholesterol and the percent free cholesterol. During the recovery period, both the absolute and percent free cholesterol decreased. Since there is an increase in total lipid mobilization including total cholesterol mobilization during exercise, it is expected that the free cholesterol level will increase as well. Since the percent free cholesterol level increases, it is evidence that the enzyme LCAT is unable to esterify the free cholesterol at the same rate that the free cholesterol

is being mobilized. A study by Abdulla et al (1) has noted that free cholesterol is mobilized faster than saturated or monosaturated cholesterol esters which supports our results.

CONCLUSIONS

Within the limitations of this study, the data suggest the following:

1. There is no significant change in serum free cholesterol level due to a three-month physical fitness program.
2. The high-fit group had a significantly lower free cholesterol level than the low-fit group.
3. There was no significant differences between fitness groups for the percent free cholesterol.
4. There were no significant differences between age groups for the free cholesterol or percent free cholesterol.
5. Acute physical exercise increases both the absolute and percent free cholesterol from the resting state to the submaximal exercise state, to the maximum exercise state. There is a decrease in both the absolute and percent free cholesterol in the recovery period.

Table 1. Means and Standard Deviations of the Selection Variables for the four groups on the Pre and Post Physical Fitness Program Tests

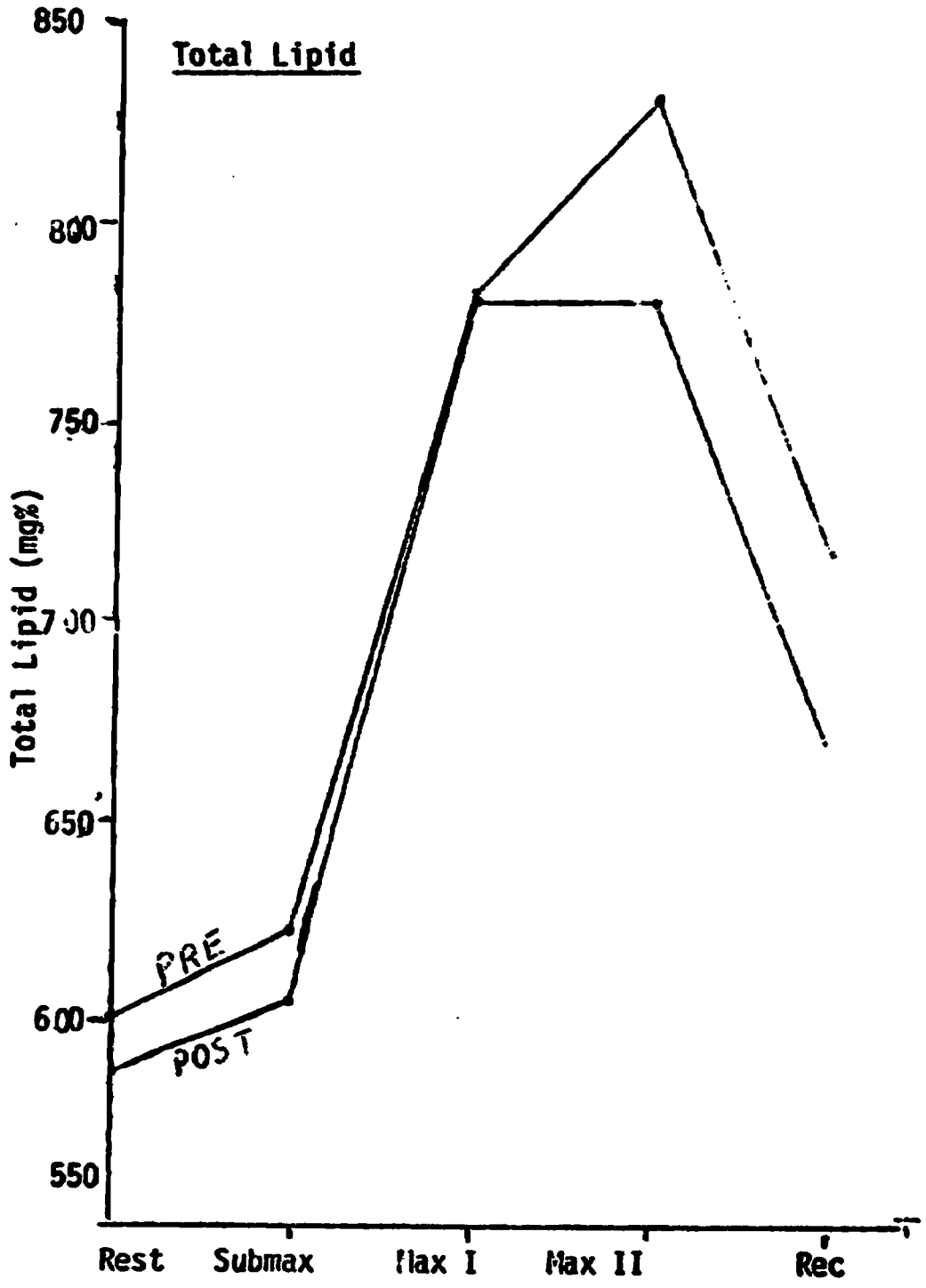
Variable	Fitness Group	Age Group	Pre Test $\bar{X}$	Pre Test S.D.	Post Test $\bar{X}$	Post Test S.D.
Age						
	Low	Young	37.2	2.6		
	Low	Old	53.2	5.5		
	High	Young	36.2	8.3		
	High	Old	52.5	4.2		
Weight						
	Low	Young	181.8	).3	179.7	23.3
	Low	Old	171.3	20.7	169.0	21.1
	High	Young	174.9	24.9	173.3	23.6
	High	Old	167.6	21.5	167.2	24.7
Physical Fitness Score						
	Low	Young	304.3	16.4	338.5	28.6
	Low	Old	288.3	62.0	336.2	53.7
	High	Young	398.7	27.9	409.8	24.5
	High	Old	411.0	40.5	431.5	27.9
Resting Serum Glucose						
	Low	Young	88.5	2.4	92.7	7.5
	Low	Old	95.0	7.5	90.8	12.1
	High	Young	91.2	5.3	85.8	5.6
	High	Old	87.8	8.8	91.3	5.5
Resting Serum Triglyceride						
	Low	Young	164.8	80.5	134.3	50.7
	Low	Old	153.3	88.5	149.5	64.0
	High	Young	82.3	29.6	89.2	30.4
	High	Old	98.2	22.8	94.7	28.9

Table 2: Means and Standard Deviations of the Serum Total Lipid Level and the Serum Total Cholesterol Level for the Total Group on the Pre and Post Test.

	Total Lipid			
	Pre		Post	
Metabolic Stress	$\bar{X}$	S.D.	$\bar{X}$	S.D.
Rest	586.1	119.0	601.3	134.0
Submax Exercise	605.5	147.1	624.8	158.4
Max I Exercise	781.8	150.0	784.6	146.9
Max II Exercise	781.8	150.0	833.5	187.0
Recovery	672.3	144.2	720.5	156.2

	Total Cholesterol			
	PRE		Post	
Metabolic Stress	$\bar{X}$	S.D.	$\bar{X}$	S.D.
Rest	236.8	45.9	232.0	52.5
Submax Exercise	246.9	46.3	250.1	55.8
Max I Exercise	271.2	48.6	273.5	59.4
Max II Exercise	271.2	48.6	276.3	58.6
Recovery	239.5	46.0	245.3	51.3

Figure 1: Mean Serum Total Lipid and the Total Cholesterol Assessed at Five Stages of Metabolic Stress on the Pre and Post Tests



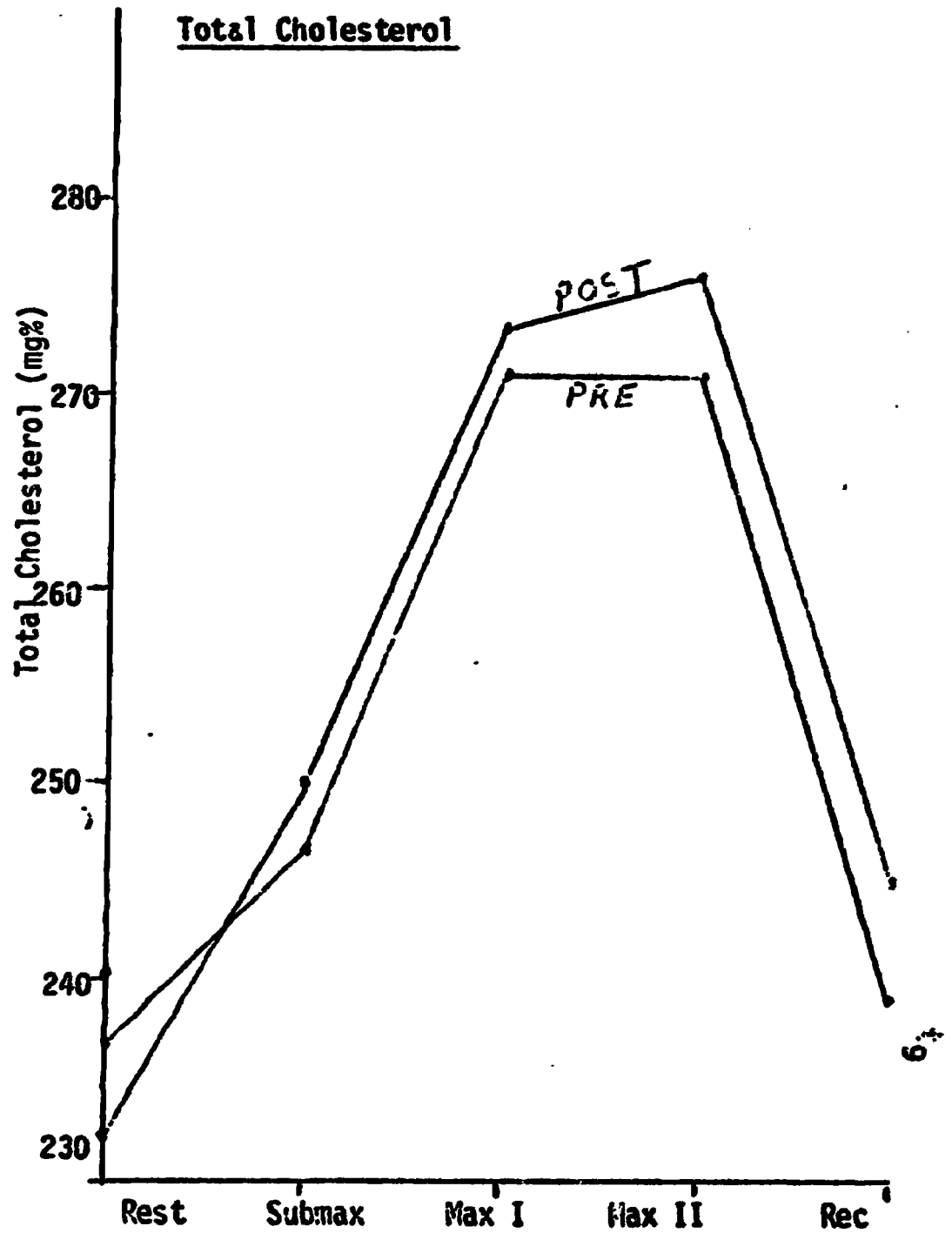


Table 3: ANOVA Results from the four test groups on the Serum Total Lipid and Serum Total Cholesterol Concentration.

				Total Lipid	Total Cholesterol
Variable		d.f.	test	Observed F	Observed F
I	Fitness	1	K(IJ)	3.64*	2.30
J	Age	1	K(IJ)	0.23	0.29
L	Time (pre vs Post)	1	KL(IJ)	1.41	0.01
M	Stress	4	KM(IJ)	125.77***	122.59***
IJ	Fitness x Age	1	K(IJ)	1.39	1.45
IL	Fitness x Time	1	KL(IJ)	0.05	0.15
JL	Age x Time	1	KL(IJ)	0.33	0.84
IM	Fitness x Stress	4	KM(IJ)	3.12*	2.25*
JM	Age x Stress	4	KM(IJ)	1.25	0.53
LM	Time x Stress	4	KLM(IJ)	1.49	2.14*
K(IJ)	Subjects within groups	20	None	----	------
IJL	Interaction	1	KL(IJ)	0.11	0.34
IJM	Interaction	4	KM(IJ)	0.62	1.79
ILM	Interaction	4	KLM(IJ)	1.34	0.19
JLM	Interaction	4	KLM(IJ)	0.23	0.38
KL(IJ)	Interaction	20	None	----	------
KM(IJ)	Interaction	80	None	----	------
IJLM	Interaction	4	KLM(IJ)	0.31	1.03
KLM(IJ)	Interaction	80	-------	----	------
Mean		1			
Total		240			

*Significant at the .90 level
**Significant at the .95 level
***Significant at the .99 level

Table 4: Means and Standard Deviations of the Serum Free Cholesterol Level and the % Free Cholesterol for the Total Group on the Pre and Post Tests.

	Free Cholesterol			
	Pre		Post	
Metabolic Stress	$\bar{X}$	S.D.	$\bar{X}$	S.D.
Rest	56.6	11.6	53.9	13.5
Submax Exercise	60.9	11.9	60.5	15.5
Max I Exercise	68.6	13.3	67.4	16.1
Max II Exercise	68.6	13.3	67.3	15.4
Recovery	59.9	13.4	58.7	14.5

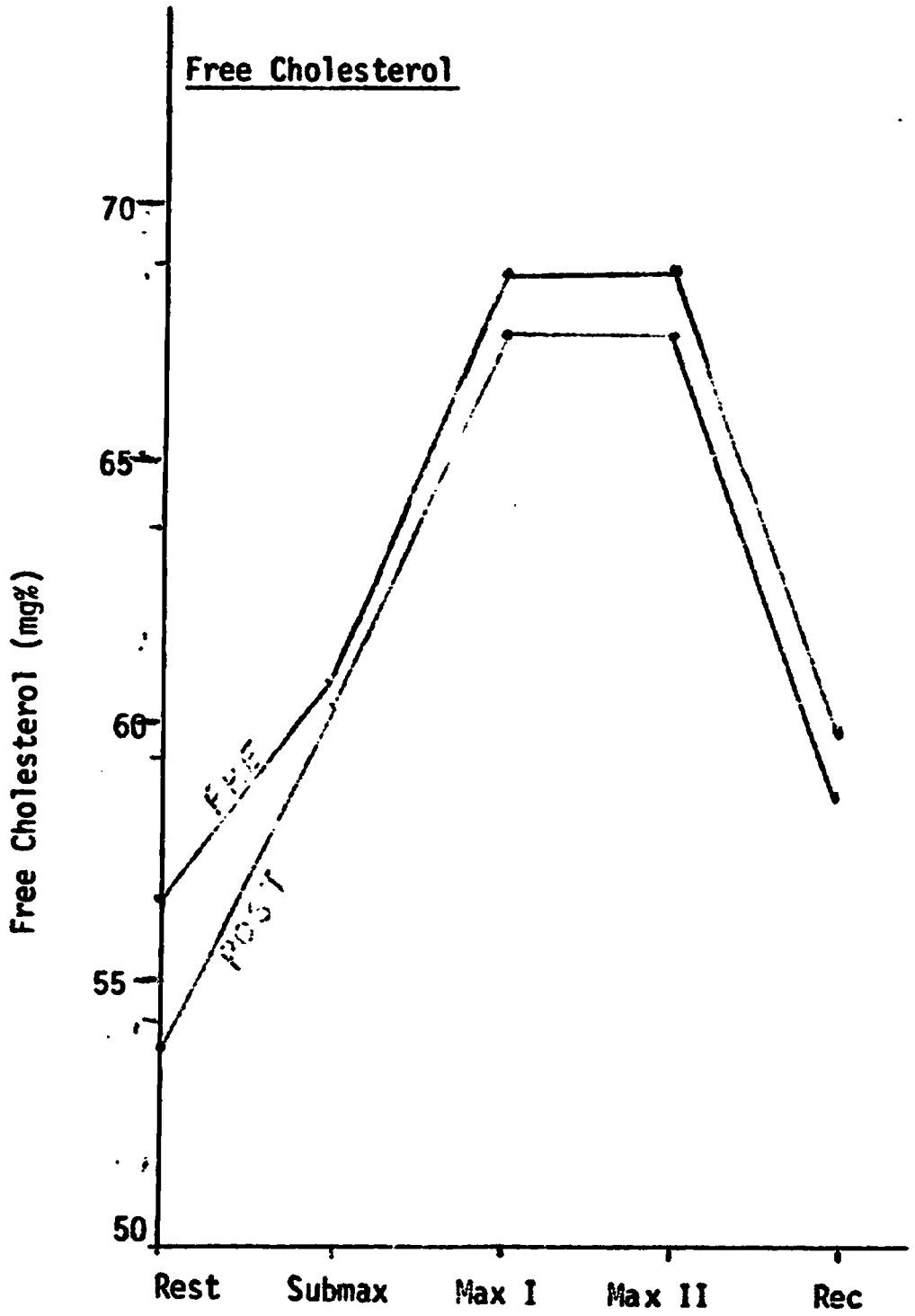
	% Free Cholesterol			
	Pre		Post	
Metabolic Stress	$\bar{X}$	S.D.	$\bar{X}$	S.D.
Rest	24.0	2.6	23.1	2.1
Submax Exercise	24.8	3.1	24.1	2.1
Max I Exercise	25.4	3.2	24.6	2.3
Max II Exercise	25.4	3.2	24.4	2.3
Recovery	24.9	2.9	23.9	2.3

Table 5: Observed F Values of the ANOVA from the Four Test Groups on the Serum Free Cholesterol and the % Free Cholesterol Concentrations

Variable		df	Test	Free Cholesterol Observed F	% Free Cholesterol Observed F
I	Fitness	1	K(IJ)	3.67*	1.76
J	Age	1	K(IJ)	0.22	0.02
L	Time	1	KL(IJ)	0.37	1.60
M	Stress	4	KM(IJ)	82.63***	7.43***
IJ	Fitness x Age	1	K(IJ)	1.51	0.03
IL	Fitness x Time	1	KL(IJ)	0.57	0.29
JL	Age x Time	1	KL(IJ)	0.01	0.11
IM	Fitness x Stress	4	KM(IJ)	0.10	0.39
JM	Age x Stress	4	KM(IJ)	0.38	1.16
LM	Time x Stress	4	KLM(IJ)	0.61	0.10
K(IJ)	Subjects within Groups	20	None	-----	----
IJL	Interaction	1	KL(IJ)	0.01	0.11
IJM	Interaction	4	KM(IJ)	1.07	0.69
ILM	Interaction	4	KLM(IJ)	0.47	0.59
JLM	Interaction	4	KLM(IJ)	0.51	0.78
KL(IJ)	Interaction	20	None	----	----
KM(IJ)	Interaction	80	None	----	----
IJLM	Interaction	4	KLM(IJ)	0.78	0.26
KLM(IJ)	Interaction	80	-------	----	----
Mean		1			
Total		240			

*Significant at the .90 Level
**Significant at the .95 Level
*** Significant at the .99 Level

Figure 2: Mean Serum Free Cholesterol and % Free Cholesterol Assessed at Five Stages of Metabolic Stress for Pre and Post Physical Fitness Tests.



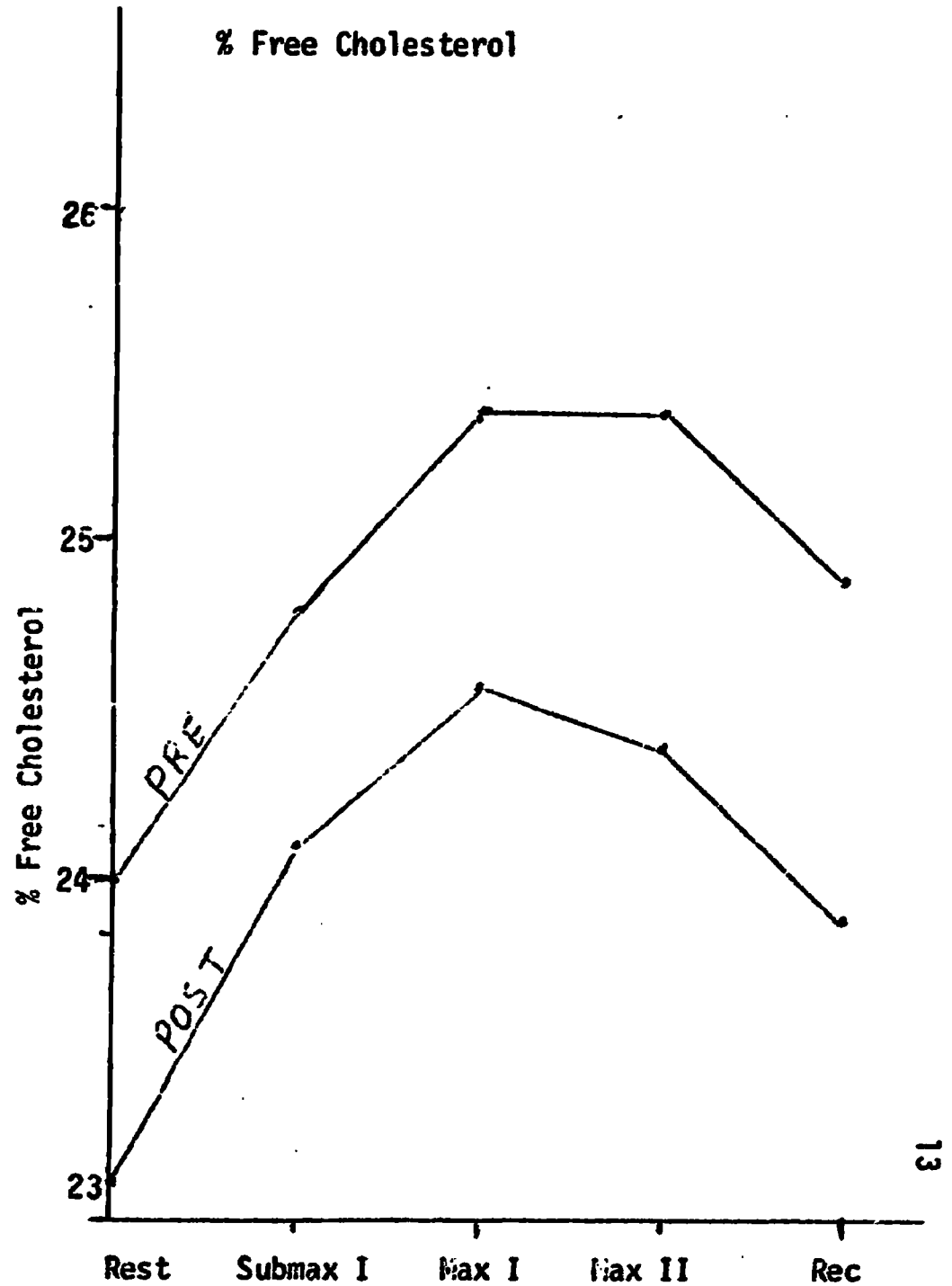
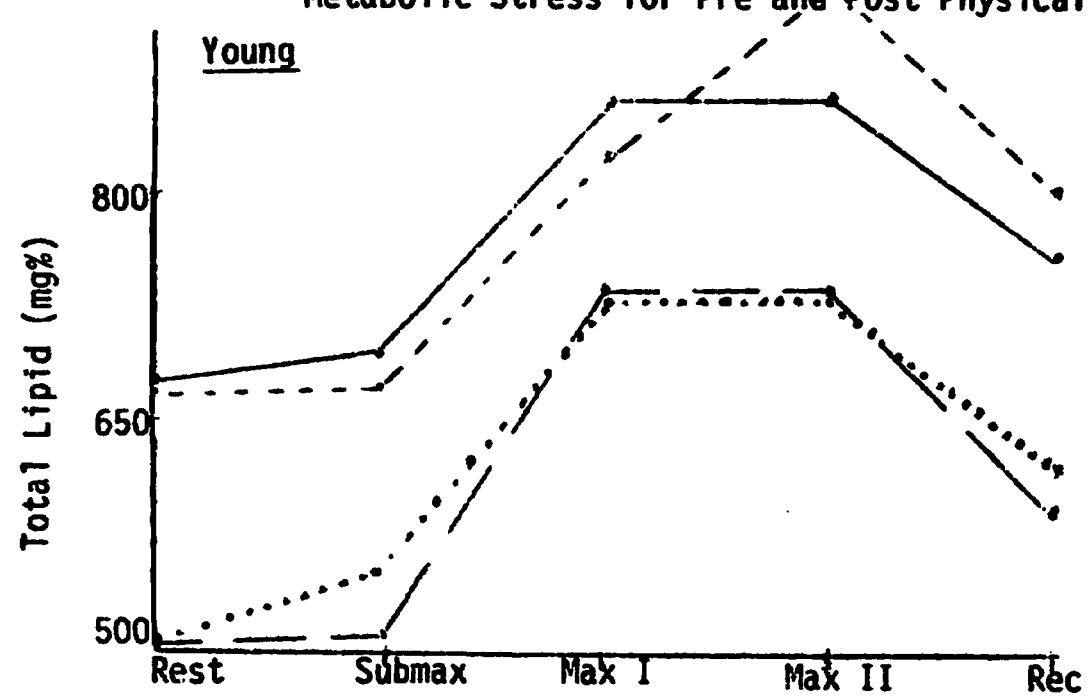
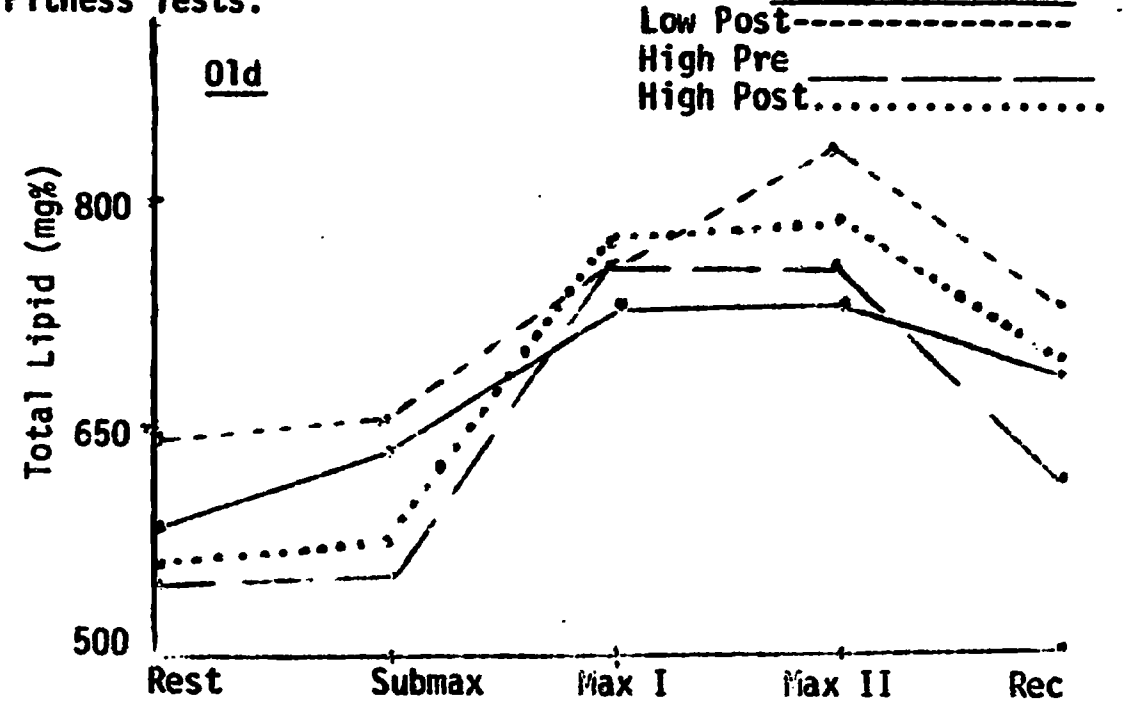


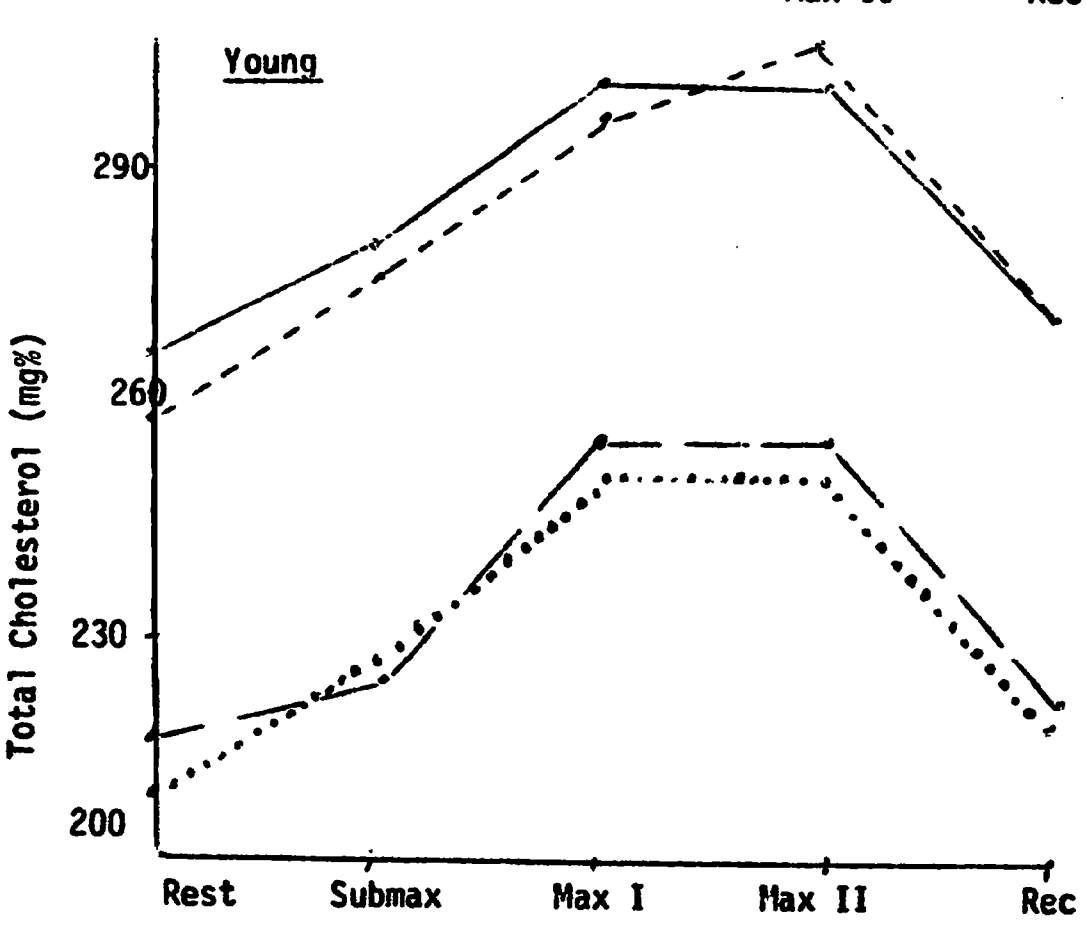
Table 6: Means and Standard Deviations of the Serum Total Lipid Level and the Serum Total Cholesterol Level for the Four Sub-groups and the Total Group on the Pre and Post Tests.

			Total Lipid				Total Cholesterol			
			Pre		Post		Pre		Post	
Metabolic Stress	Fitness Group	Age Group	X	S.D.	X	S.D.	X	S.D.	X	S.D.
Rest	Total - Group		586.1	119.0	601.3	134.0	236.8	45.9	232.0	52.5
	Low	Young	684.5	157.3	683.3	152.1	269.0	66.3	260.3	84.0
	Low	Old	591.5	73.5	647.8	149.0	241.0	29.6	239.8	37.2
	High	Young	509.7	105.1	506.8	97.1	216.3	41.3	207.8	43.4
	High	Old	558.1	66.0	567.3	71.1	220.7	24.9	219.8	20.4
Submax Exercise	Total - Group		605.5	147.1	624.8	158.4	246.9	46.3	250.1	55.8
	Low	Young	705.3	186.8	687.5	234.9	282.3	63.3	279.7	89.1
	Low	Old	639.7	132.5	662.0	154.4	246.0	33.0	249.0	38.9
	High	Young	515.3	118.2	566.2	95.0	224.5	39.9	226.0	50.1
	High	Old	561.7	87.9	583.7	121.7	234.8	30.0	245.8	24.5
Max I Exercise	Total - Group		781.8	150.0	784.6	146.9	271.2	48.6	273.5	59.4
	Low	Young	876.2	202.0	836.8	190.6	304.3	75.5	301.0	99.2
	Low	Old	737.5	159.1	776.3	149.1	265.3	35.3	268.0	41.7
	High	Young	750.0	89.7	745.8	151.7	256.5	35.4	251.2	47.8
	High	Old	763.7	121.4	779.3	112.1	258.7	29.0	274.0	29.2
Max II	Total - Group		781.8	150.0	833.5	187.0	271.2	48.6	276.3	58.6
	Low	Young	876.2	202.0	951.0	284.4	304.3	75.5	310.2	93.5
	Low	Old	737.5	159.1	839.5	169.8	265.3	35.3	265.5	42.2
	High	Young	750.0	89.7	750.7	133.0	256.5	35.4	251.8	49.0
	High	Old	763.7	121.4	792.8	81.7	258.7	29.0	277.5	25.4
Recovery	Total - Group		672.3	144.2	720.5	156.2	239.5	46.0	245.3	51.3
	Low	Young	774.0	182.8	817.5	241.5	274.7	67.0	274.3	83.2
	Low	Old	687.5	165.6	735.8	103.8	237.5	31.5	243.0	34.1
	High	Young	601.7	82.2	639.5	136.8	221.5	32.1	218.2	39.7
	High	Old	626.0	82.8	689.2	61.6	224.2	32.6	245.5	23.3

Figure 3: Mean Serum Total Lipid and Total Cholesterol Assessed at Five Stages of Metabolic Stress for Pre and Post Physical Fitness Tests.







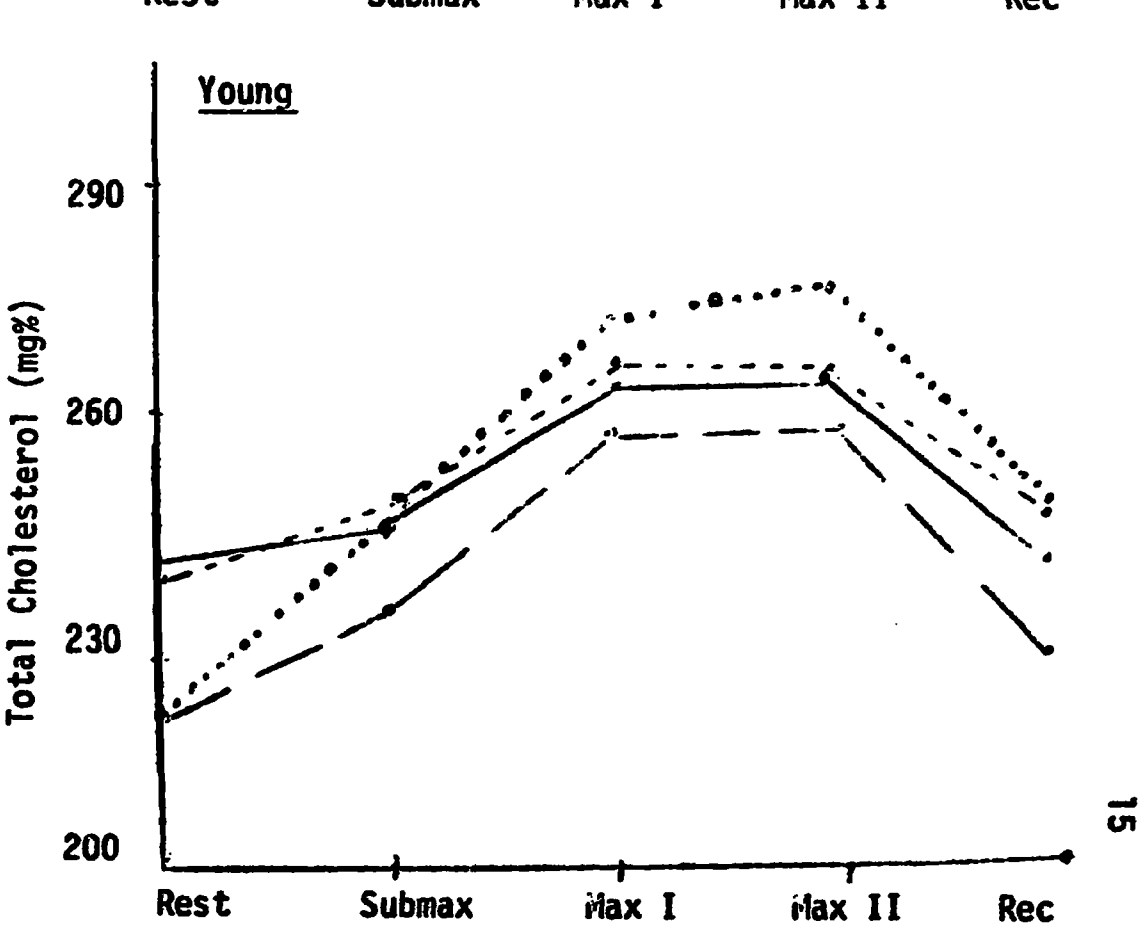
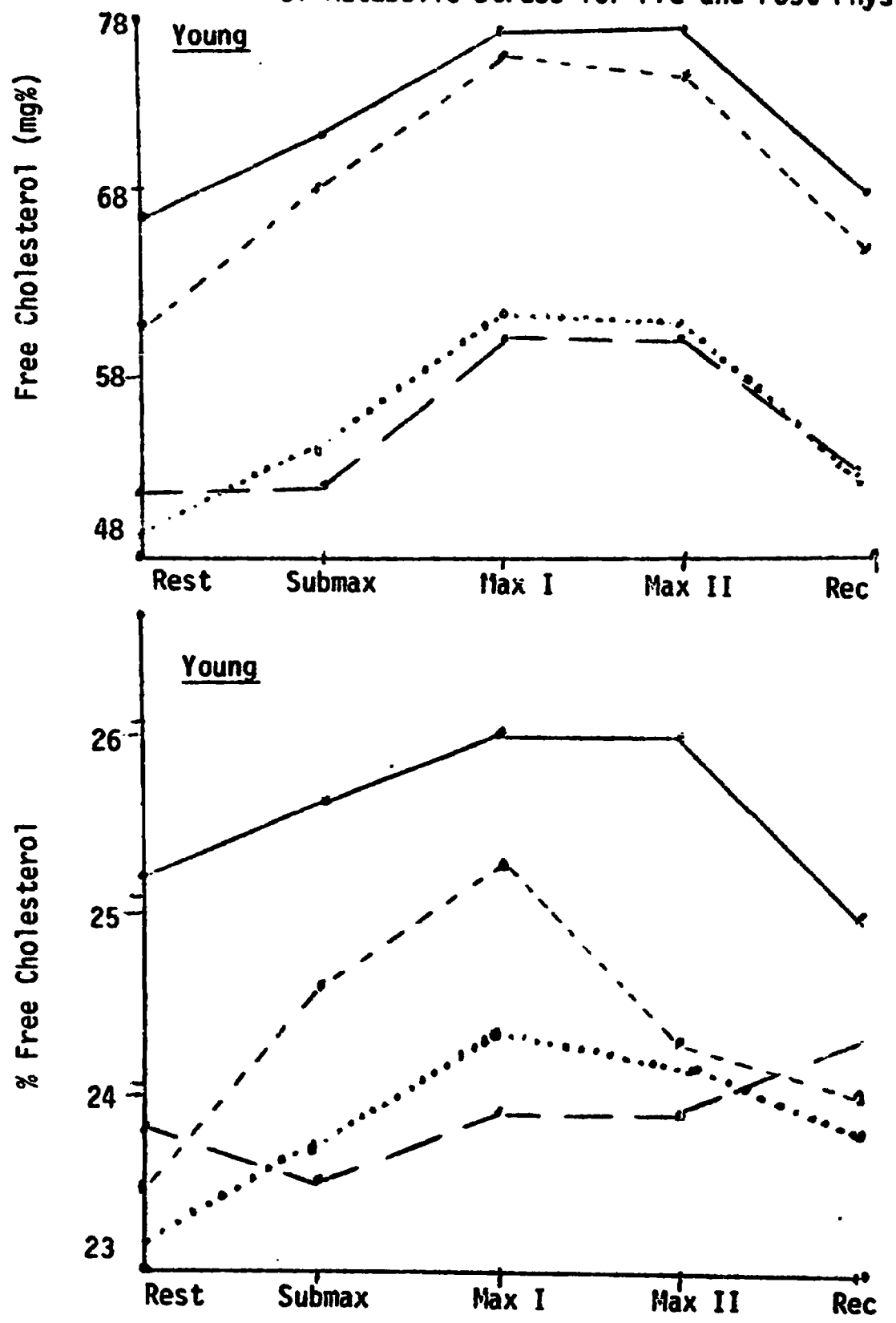


Table 7: Means and Standard Deviations of the Free Cholesterol Level and the % Free Cholesterol for the Four Sub-groups and the Total Group on the Pre and Post Tests.

Metabolic Stress	Fitness Group	Age Group	Free Cholesterol				% Free Cholesterol			
			Pre		Post		Pre		Post	
			X̄	S.D.	X̄	S.D.	X̄	S.D.	X̄	S.D.
Rest	Total - Group		56.6	11.6	53.9	13.5	24.0	2.6	23.1	2.1
	Low	Young	66.8	12.8	61.0	20.1	25.2	2.3	23.4	1.9
	Low	Old	56.5	10.1	56.2	8.1	23.5	3.5	23.6	2.2
	High	Young	51.2	9.2	49.2	14.5	23.8	2.6	23.1	2.5
	High	Old	52.0	9.1	49.3	6.3	23.4	2.0	22.4	2.3
Submax Exercise	Total - Group		60.9	11.9	60.5	15.5	24.8	3.1	24.1	2.1
	Low	Young	71.3	11.0	69.0	23.0	25.6	2.6	24.6	2.4
	Low	Old	62.2	11.1	59.8	12.3	25.4	3.3	23.9	1.9
	High	Young	52.2	9.3	54.0	16.1	23.5	4.3	23.7	3.2
	High	Old	57.8	9.2	59.3	5.4	24.6	2.1	24.2	0.9
Max I Exercise	Total - Group		68.6	13.3	67.4	16.1	25.4	3.2	24.6	2.3
	Low	Young	77.8	14.3	76.2	25.3	26.0	3.2	25.3	2.1
	Low	Old	70.7	14.9	65.5	11.4	26.7	4.4	24.5	2.4
	High	Young	60.8	7.9	61.8	16.0	23.9	3.3	24.4	3.2
	High	Old	65.0	10.4	66.0	6.0	25.0	1.4	24.2	1.8
Max II Exercise	Total - Group		68.6	13.3	67.3	15.4	25.4	3.2	24.4	2.3
	Low	Young	77.8	15.3	75.7	24.6	26.0	3.2	24.3	1.8
	Low	Old	70.7	14.9	66.0	9.4	26.7	4.4	25.0	2.6
	High	Young	60.8	7.9	61.2	15.8	23.9	3.3	24.2	3.2
	High	Old	65.0	10.4	66.3	4.2	25.0	1.4	24.0	1.8
Recovery	Total - Group		59.9	13.4	58.7	14.5	24.9	2.9	23.9	2.3
	Low	Young	68.7	19.0	65.5	23.7	25.0	2.0	24.0	3.2
	Low	Old	60.7	11.8	59.3	7.1	25.6	3.5	24.5	1.9
	High	Young	53.0	5.2	52.7	14.9	24.3	3.9	23.8	2.8
	High	Old	56.0	11.2	57.2	4.9	24.9	2.4	23.3	1.6

Figure 4: Mean Serum Free Cholesterol and % Free Cholesterol Assessed at Five Stages of Metabolic Stress for Pre and Post Physical Fitness Tests.



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